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(54) Title: COMPOSITIONS INCLUDING R-NSAIDS AND THERAPEUTIC AND PROPHYLACTIC METHODS EMPLOYING SAID COMPOSITIONS

(57) Abstract

A composition including an enantiomerically stable R-NSAID or a pharmaceutically acceptable salt thereof, the composition being substantially free of the S-enantiomer of the selected R-NSAID.

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COMPOSITIONS INCLUDING R-NSAIDS AND THERAPEUTIC AND PROPHYLACTIC METHODS EMPLOYING SAID COMPOSITIONS

This is a continuation-in-part of U.S. Patent Application Serial No. 08/706,634, filed September 6, 1996, which in turn was a continuation-in-part of U.S. Patent Application Serial No. 08/402,797, filed March 13, 1995, both of which are incorporated in their entireties herein by reference.

Field of the Invention

The present invention relates to compositions and methods useful in the treatment and prevention of neoplastic diseases, such as colorectal and other gastrointestinal epithelial cancers as well as breast cancer and other cancers, and also useful in the treatment of cystic fibrosis and the prevention or delay of onset of symptoms of Alzheimer's Disease.

Background of the Invention

Cancer of the colon is common in the western world and is an important cause of morbidity and mortality, having an incidence of about 5% in the U.S. population. As with other types of cancers, cancers of the gastrointestinal tract, including colon cancer, are characterized by abnormal development in cell proliferation and differentiation in the gastrointestinal tract.

The gastrointestinal tract, including the rectum and colon, is lined with epithelial cells which have a high proliferation rate. The lining of the colon, in particular, made up of columnar rows of epithelial cells, is characterized by a series of indentations or crypts. Epithelial cells in the bottom regions of the crypts proliferate and move upward toward the tops of the crypts. In the normal colon, the proliferation region of the large intestine normally occupies the basal or deeper three-quarters of the crypts. A relationship has been observed between the expansion of cell proliferation zones to the upper regions of the crypts and colon cancer. *See* M. Lipkin, "Biomarkers of Increased Susceptibility to Gastrointestinal Cancer: New Application to Studies of Cancer Prevention in Human Subjects," *Cancer Research*, Vol. 48, pp. 235-245 (January 15, 1988).

More generally, neoplastic diseases are conditions in which abnormal proliferation of cells results in a mass of tissue called a neoplasm or tumor. Neoplasms have varying degrees of abnormalities in structure and behavior. Some

neoplasms are benign while others are malignant or cancerous. An effective treatment of neoplastic disease would be considered a valuable contribution to the search for cancer preventive or curative procedures.

There has been an intensive search for chemoprotective agents for all individuals at risk for colon cancer and other gastrointestinal cancers, particularly individuals over the age of 45. One class of potentially therapeutically useful compounds are the non-steroidal antiinflammatory drugs ("NSAIDs"). NSAIDs, presently in common use as anti-inflammatory agents and as analgesics, are known to have neoplasia chemoprevention and other anti-neoplastic benefits. Physiologically, NSAIDs are known to inhibit the biosynthesis of prostaglandins by the inhibition of the cyclooxygenase enzyme which is ubiquitous in mammalian tissues. *See* Buckley *et al.*, *Drugs*, 39(1):86-109 (1990). The role of NSAIDs in prevention of colorectal cancer is discussed in Heath *et al.*, "Nonsteroidal Antiinflammatory Drugs and Human Cancer," *Cancer*, Vol. 74, No. 10, pp. 2885-2888 (November 15, 1994).

However, the use of NSAIDs in colon cancer prevention has been associated with severe undesirable side effects, which include gastrointestinal, renal and hepatic toxicities, as well as increases in bleeding times due to disruption of platelet function (*e.g.*, thrombocytopenia), and prolongation of gestation due to uterine effects. Another serious side effect associated with the use of certain NSAIDs is leukopenia (decreased white cell count in the blood), and consequent agranulocytosis.

Agranulocytosis is a life-threatening condition that develops very rapidly, and that is difficult to detect even with periodic white-cell counts. The leukopenia/agranulocytosis syndrome has been described for several NSAIDs, such as indomethacin, ketoprofen, and ibuprofen. Indeed, such NSAIDs are contraindicated in patients whose immune systems are compromised by HIV infection, chemotherapy, ionizing irradiation, corticosteroids, immunosuppressives, etc., or by such conditions as emphysema, bronchiectasis, diabetes mellitus, leukemia, burns and the like. A recent review of the adverse effects of NSAIDs is Borda et al., "NSAIDs: A Profile of Adverse Effects," Hanley and Belfus, Inc., Philadelphia, PA, 1992.

The most recent epidemiologic survey showing that both aspirin and NSAIDs confer protection against colon cancer is Peleg, *et al.*, "Aspirin and Nonsteroidal Anti-inflammatory Drug Use and the Risk of Subsequent Colorectal Cancer," Arch.

Intern. Med., Vol. 154, pp. 394-400 (February 28, 1994). This reference identifies a causal relationship between the use of NSAIDs, such as indomethacin, sulindac and peroxicam, and prevention of cancer of the large bowel and rectum. A risk benefit analysis is suggested, however, due to the severe potential gastrointestinal and renal side effects, particularly in the elderly.

The standard treatment for colon cancer currently consists of the administration of a known cancer fighting agent, 5-fluorouracil in combination with the antibiotic levamisole. No improvement in survival among colon cancer patients was shown when 5-fluorouracil was administered alone. The addition of levamisole, which is known to stimulate the immune system and increase T-cell count, showed improved survival rate among these patients. See Moertel et al., "Levamisole and Fluorouracil for Adjuvant Therapy of Resected Colon Carcinoma," N Engl J Med 1990; 322:352-358.

Many NSAIDs exhibit molecular chirality, and thus have R- and S-enantiomers. Such compounds typically are produced as racemic mixtures, which can subsequently be separated into the individual enantiomers.

The enantiomers of several 2-arylpropionic acid NSAIDs are discussed in Yamaguchi *et al.*, *Nippo Yakurigaku Zasshi,* 90:295-302 (1987). Yamaguchi *et al.* state that the S-enantiomers of 2-arylpropionic acids have 15-300 times higher prostaglandin synthetase inhibitory activities than the R-enantiomers in the rat.

Caldwell *et al.*, *Biochem. Pharmacol.* 37: 105-114 (1988) allege that "at best, the R-isomers [of 2-arylpropionic acids] function as prodrugs for the therapeutically active S-forms" when the racemic drug is administered and thus add to both in the therapeutic and toxic effects of the active S-enantiomers. Caldwell *et al.* further contend that "at worst, the R-enantiomers are undesirable impurities in the active drug" causing difficulties due to non-stereoselective toxicity. The authors indicate that the use of the S-isomers alone should provide safer and more effective use of this class of drugs.

Similarly, it has been generalized that the pharmacokinetics of the enantiomers of 2-arylpropionic acids are different due, at least in part, to the unidirectional metabolic inversion of the R- to the S-enantiomer. However, it has been found that this interconversion depends on the particular compound and the particular species in which it is administered. Jamali, *Eur. J. Drug Metabolism Pharmaco.* 13: 1-9(1988).

Because of the toxicity and side effects previously described, many NSAIDs

are no longer in use in human medicine as analgesics. Some of these NSAIDs include tiaprofenic acid, suprofen, carprofen, pirprofen and indoprofen.

A need has been identified for new formulations of NSAIDs that are effective in treating colorectal and other cancers but are more tolerable with regard to gastrointestinal toxicity. Thus, it would be particularly desirable to provide compositions and methods for the prevention of neoplasia and colorectal cancer but without the aforementioned disadvantages.

Another disease for which effective treatment is needed is cystic fibrosis. Cystic fibrosis (CF) is a heritable disease that follows an autosomal recessive pattern of transmittance. It is the most common lethal genetic disease in the United States. The approximate frequency in Caucasians is 1 in 2000. Cystic fibrosis is characterized by abnormal eccrine and exocrine gland function. In particular, mucous glands produce viscous secretions which lead to chronic pulmonary disease, insufficient pancreatic and digestive function and abnormally concentrated sweat. The most prominent theories of CF etiology focus on alterations in physiochemic properties of exocrine secretions, the regulation of exocrine gland secretions, electrolyte transport and abnormalities in serum. Typical presentations include early onset of respiratory symptoms such as colds, and recurrent respiratory infections later in life. CF patients show evidence of decreasing pulmonary function with time, and their sputum cultures often display *S. aureus, P. aeruginosa* and *P. capacia*.

The major source of CF morbidity is pulmonary disease. More than 98% of CF patients die of either respiratory failure or pulmonary complications. Antibiotics are the key element in increasing survival. Prior to the 1950's, when modern antibiotics began to become available, patients typically survived for only a few years. At present, the medial survival age is 24. Consequently, stimulation of neutrophil function as a means of clearing bacterial foci is thought to be an appropriate focus of treatment.

It has been reported (M.W. Konstan et al., *New England J. Med.* 1995; 332:848-854) that high doses of racemic ibuprofen in cystic fibrosis patients over a four-year period slows progression of the lung disease. However, gastrointestinal side effects due to the presence of *S*(+) ibuprofen severely limit the chronic use of this therapy, particularly at high dose and as the racemate (see Wechter, W.J. *J. Clin. Pharmacol.* 1994; 34:1036-1042 and Wechter et al. *Chirality* 2993; 5:492-494). It is believed that high doses of racemic ibuprofen

inhibits the influx of neutrophils to the alveolar crevices, while low doses increase the influx of neutrophils. The high doses employed in the Konstan study also appear to cause conjunctivitis and epistaxis.

Still another disease for which effective treatment is needed is Alzheimer's Disease (AD) is a degenerative brain disorder associated with extensive loss of specific neuronal subpopulations and characterized clinically by progressive loss of memory, cognition, reasoning, judgment and emotional stability that gradually leads to profound mental deterioration and ultimately death. AD is a common cause of progressive mental failure (dementia) in aged humans and is believed to represent the fourth most common medical cause of death in the United States. AD has been observed in varied races and ethnic groups worldwide and presents a major present and future public health problem. The disease is currently estimated to affect up to four million individuals in the United States alone. To date, AD has proven to be incurable, and presently causes up to 100,000 deaths yearly.

The brains of individuals with AD exhibit neuronal degeneration and characteristic lesions variously referred to as amyloidogenic plaques, vascular amyloid angiopathy, and neurofibrillary tangles. Large numbers of these lesions, particularly amyloidogenic plaques and neurofibrillary tangles, are generally found in several areas of the human brain important for memory and cognitive function in patients with AD. Smaller numbers of these lesions in a more restricted anatomical distribution are found in the brains of most aged humans who do not have clinical AD, as well as patients suffering from Down's Syndrome and Hereditary Cerebral Hemorrhage with Amyloidosis of the Dutch-Type.

It is presently believed that progressive cerebral deposition of particular amyloidogenic proteins, beta -amyloid proteins (beta AP), play a seminal role in the pathogenesis of AD and can precede cognitive symptoms by years or decades. Recently, it has been shown that beta AP is released from neuronal cells grown in culture and is present in cerebrospinal fluid (CSF) of both normal individuals and AD patients.

A possible correlation to the plaque pathology has been developed by several groups demonstrating the direct beta AP neurotoxicity toward cultured neurons. More recently, in addition to the direct neurotoxicity, an inflammatory response in the AD brain, perhaps elicited by beta AP, also contributes to the pathology of the disease. A limited clinical trial with the NSAID indomethacin exhibited a

retardation in the progression of Alzheimer's dementia (Rogers et al., Science, 260:1719-1720 (1993)).

Previous methods of treating AD are disclosed, for example, in U.S. Patent No. 5,576,353 (use of N-propargyl-aminoindan compounds) and U.S. Patent No. 5,552,415 (use of raloxifene and related compounds). A continuing need exists for effective methods for preventing, delaying, and treating AD.

Summary of the Preferred Embodiments

In accordance with one aspect of the present invention, a composition useful in preventing colorectal cancer includes an enantiomerically stable R-NSAID or a pharmaceutically acceptable salt thereof in an amount effective to elicit a chemoprotective effect. The composition is substantially free of the S-enantiomer of the R-NSAID.

In a preferred embodiment, the R-NSAID is a propionic acid derivative, particularly preferably R-flurbiprofen.

According to another aspect of the present invention, a method of eliciting a colorectal chemoprotective effect in a mammal with reduced gastrointestinal toxicity includes the step of administering to the mammal a composition as described above.

In accordance with still another aspect of the present invention, a method of treating a neoplastic disease in a mammal with reduced gastrointestinal toxicity includes the step of administering to the mammal a composition as described above.

In accordance with yet another aspect of the present invention, a method of treating cystic fibrosis is provided comprising the step of administering to a patient in need of such treatment a composition comprising an effective cystic fibrosis therapeutic amount of an enantiomerically stable R-NSAID or a pharmaceutically acceptable salt thereof. The composition is substantially free of the S-enantiomer of said R-NSAID.

In accordance with another aspect of the present invention, there is provided a composition comprising an effective cystic fibrosis therapeutic amount of an enantiomerically stable R-NSAID or a pharmaceutically acceptable salt thereof, said composition being substantially free of the S-enantiomer of said R-NSAID.

In accordance with a further aspect of the present invention, a method of preventing or delaying the onset of Alzheimer's Disease is provided comprising the step of administering to a patient in need of such treatment a composition

comprising an effective Alzheimer's Disease prophylactic amount of an enantiomerically stable R-NSAID or a pharmaceutically acceptable salt thereof. The composition is substantially free of the S-enantiomer of said R-NSAID.

In accordance with yet a further aspect of the present invention, there is provided a composition comprising an effective Alzheimer's Disease prophylactic amount of an enantiomerically stable R-NSAID or a pharmaceutically acceptable salt thereof, said composition being substantially free of the S-enantiomer of said R-NSAID.

Other objects, features and advantages of the present invention will become apparent to those skilled in the art from the following detailed description. It is to be understood, however, that the detailed description and specific examples, while indicating preferred embodiments of the present invention, are given by way of illustration and not limitation. Many changes and modifications within the scope of the present invention may be made without departing from the spirit thereof, and the invention includes all such modifications.

Detailed Description of the Preferred Embodiments

It has surprisingly been discovered that enantiomerically stable R-isomers of NSAIDs are highly effective in eliciting a colorectal chemoprotective effect, and are also useful in treating neoplastic disease, such as adenocarcinomas, including but not limited to colon, rectal, breast, lung and prostate cancers. Prophylactic and/or therapeutic administration of compositions including R-NSAIDs in substantially pure form (that is, substantially free of the S-enantiomer of the selected NSAID) is accompanied by a significant reduction in adverse effects associated with the administration of S-enantiomers or racemic mixtures of NSAIDs. Such adverse effects include, but are not limited to, thrombocytopenia and consequent increases in bleeding times; leukopenia and agranulocytosis; prolongation of gestation; gastrointestinal toxicities such as gastric and intestinal ulcerations and erosions; renal toxicities such as papillary necrosis and chronic interstitial nephritis; and hepatic toxicities, such as jaundice, acute hepatitis and hepatic failure.

The term "effective to elicit a chemoprotective effect" as used herein means that abnormal cell proliferation is reduced. A method of measuring cell proliferation in animals is the Labelling Index (LI). Epithelial cells of the distal colon are stained using a histologic biomarker of proliferating cells. Microscopic examination allows for quantification of the proportion of proliferating cells in the

crypts. A high proportion of proliferating cells or LI, particularly in the upper portion of the crypts, is an indicator of abnormal cell proliferation. A reduction in the LI of at least 10 to 50%, preferably at least 30% is associated with the reduction of abnormal cell proliferation. Of course, the particular R-NSAID used must be enantiomerically stable in the animal species being tested.

Chemoprevention in man and animals can also be measured by the inhibition of the conversion of the intestinal polyps, in an animal prone to polyposis, to neoplastic or cancerous legions.

A min/+ mouse model can also be used to measure chemopreventive effect. Chemoprevention is achieved in this model if administration of the R-NSAID retards the spontaneous production of intestinal tumors in a min/+ mouse.

Another test of chemoprotection is demonstrated by the prevention of induced tumors in a carcinogen treated mouse or rat.

The inventive compositions comprise at least one enantiomerically stable R-NSAID and are substantially free of the corresponding S-NSAID. As used herein, the term "enantiomerically stable" means that at steady state there is no more than about 20% of the circulation NSAID as its S-enantiomer and preferably no more than 10% (i.e., 90% R, 10% S). A suitable measure of this ratio is obtained by evaluating the relative concentrations of the two enantiomers in the blood plasma or urine vs. time.

The rate of change of enantiomer concentration in plasma, for example, is assumed to reflect quantitatively the change in drug concentrations throughout the body. This rate can be approximated by first-order kinetics. See Gibaldi *et al. Pharmacokinetics*, (1982) Chapter 1, pp. 1-5, which is hereby incorporated by reference.

Pharmacokinetic data and an explanation of the present state of knowledge for many NSAIDs are presented in Jamali, *"Pharmacokinetics of Enantiomers of Chiral Non-steroidal Anti-inflammatory Drugs," Eur. J. Drug Metab. Pharmacokin.* (1988), Vol. 13, No. 1, pp. 1-9, which is hereby incorporated by reference.

The term "substantially free" indicates that the amount of S-NSAID, if any, present in the composition is insufficient to elicit an adverse effect in the patient to whom the composition is administered or, at most elicits an adverse effect that is tolerable to the patient and is outweighed by the beneficial effect or effects. Preferably, the inventive composition contains at least 90% by weight of a R-NSAID and 10% by weight or less of the corresponding S-NSAID, based upon the

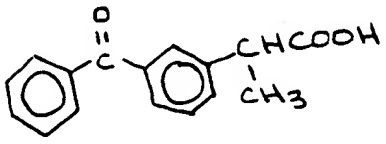
total amount of NSAID present in the composition. That is, the ratio of R-NSAID to S-NSAID in the composition is at least about 90:10. Particularly preferably, the inventive composition contains at least 99% by weight of the R-NSAID and 1% or less of the corresponding S-NSAID.

The term "eliciting a colorectal chemoprotective effect" as used herein means relieving, ameliorating or preventing colorectal cancers. Specifically, it means that abnormal cell proliferation in the colon and rectum are reduced. Measurement of these effects are as described above. Again, a reduction in the LI of at least 10 to 50%, preferably at least 30% is associated with the reduction of abnormal cell proliferation.

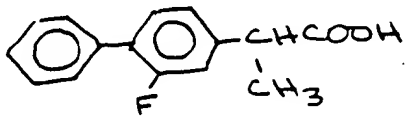
The chemical structures of NSAIDs vary. Certain NSAIDs, such as ketoprofen and flurbiprofen are arylpropionic acids, while others are cyclized derivatives of arylpropionic acids, arylacetic acids, thiazinecarboxamides, etc.. Depending on the structure of a particular NSAID, the compound may or may not exhibit chirality, *i.e*, may not have R- and S-enantiomers.

Some of the NSAIDs useful in the present invention are:

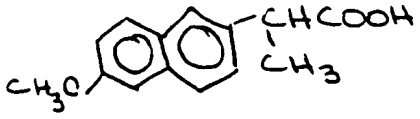
Ketoprofen



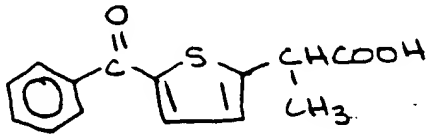
Flurbiprofen



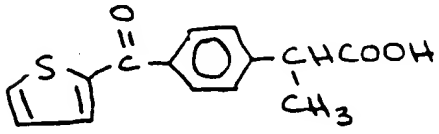
Naproxen



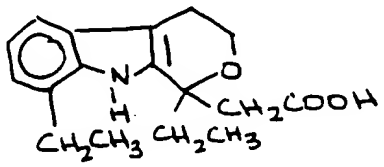
Tiaprofenic Acid



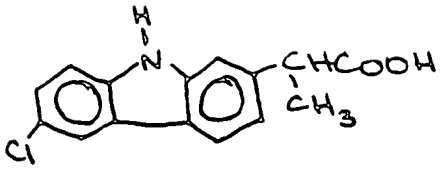
Suprofen



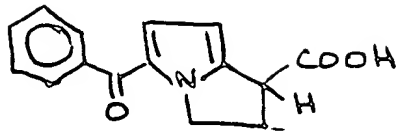
Etodolac



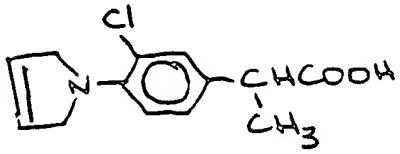
Carprofen



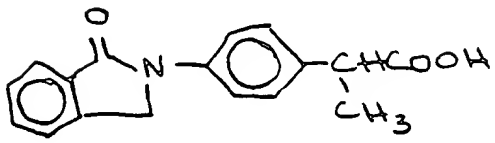
Ketorolac



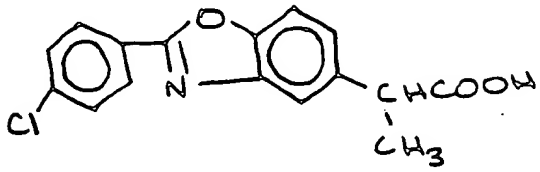
Pirprofen



Indoprofen



Benoxaprofen



In a preferred embodiment, the R-NSAID employed in the compositions and methods claimed is an arylpropionic acid, in particular a compound selected from the group consisting of R-flurbiprofen, R-ketoprofen, R-naproxen, R-tiaprofenic acid, R-suprofen, R-carprofen, R-pirprofen, R-indoprofen, and R-benoxaprofen. The R-NSAID can also be a cyclized derivative of arylpropionic acid, such as R-ketorolac, or an arylacetic acid, such as R-etodolac. All of these NSAIDs have been used in human medicine in the U.S. and/or Europe as racemates, with the exception of naproxen which is commercially available as the S-isomer only, and are enantiomerically stable. Enantiomerically unstable NSAIDs, for example propionic acid derivatives such as ibuprofen, are not encompassed by the present invention.

Descriptions of specific NSAIDs can be found in various publications. Ketoprofen, for example, is described in U.S. Patent No. 3,641,127. A description of flurbiprofen is found in U.S. Patent No. 3,755,427. Ketorolac, another chiral NSAID, is described in U.S. Patent No. 4,089,969.

A large number of NSAIDs useful according to the invention are

commercially available either in the form of racemic mixtures or as optically pure enantiomers. In all cases racemic mixtures contain equal amounts of the R- and S-isomers of the NSAID are provided. For example, the following racemates can be obtained through Sigma Chemical Co.: ketoprofen, flurbiprofen, etodolac, suprofen, carprofen, indoprofen and benoxaprofen. Naproxen, marketed as the S-isomer only, is also available from this source. Additionally, many commercial sources exist for the stereospecific R-isomers of many NSAIDs. R-ketoprofen, R-flurbiprofen and R-ketorolac, for example, are available through Sepracor, Inc.; R-naproxen can be obtained as the sodium salt through Sigma Chemical Co.; R-etodolac is available from Wyeth-Ayerst; R-tiaprofenic acid is available through Roussel (France, Canada, Switzerland, Spain, Denmark, Italy); R-suprofen is manufactured by McNiel Pharmaceuticals; R-carprofen is available from Roche; R-pirprofen is available through Ciba (France, Belgium, Denmark); R-indoprofen can be obtained through Carlo Elba (Italy, U.K.); and R-benoxaprofen is manufactured by Eli Lilly Co..

In addition to commercial sources, racemic mixtures of NSAIDs which are useful according to the invention can be produced by methods described in numerous references and U.S. Patents. Synthesis of ketoprofen, for example, is described in U.S. Patent No. 3,641,127, which is hereby incorporated by reference, while the synthesis of racemic ketorolac is disclosed in Muchowski *et al., J. Med. Chem.*, 28(8):1037-1049 (1985). The optically pure R-isomers of the selected NSAIDs can then be obtained by resolving the racemic mixtures according to well-known methods. *See, e.g.,* U.S. Patent No. 5,331,000 (R-ketoprofen) and U.S. Patent No. 5,382,591 (R-ketorolac), the contents of each of which are incorporated herein by reference.

The magnitude of a prophylactic or therapeutic dose of an R-NSAID in the acute or chronic management of cancer or neoplastic disease will vary with the particular NSAID, the severity of the condition to be treated, and the route of administration. The dose and/or the dose frequency will also vary according to the age, body weight, and response of the individual patient.

In general, the total daily dose range for a R-NSAID, for the conditions described herein, is from about 0.1 mg to about 2000 mg, in single or divided doses. Preferably, a daily dose range for cancer prevention should be between about 0.1 mg to about 500 mg in single or divided doses. The preferable daily dose for treatment of neoplastic disease (e.g., colon, rectal, breast, lung and

prostate cancers) should be about 1.0 mg to about 2000 mg in single or divided doses.

In managing the patient, the therapy should be initiated at a lower dose, perhaps about 0.1 mg to about 100 mg and increased up to about 1000 mg or higher depending on the patient's global response. It is further recommended that infants, children, patients over 65 years, and those with impaired renal or hepatic function, initially receive low doses, and that they be titrated based on individual response(s) and blood level(s).

It may be necessary to use dosages outside these ranges in some cases as will be apparent to those skilled in the art. Further, it is noted that the ordinary skilled clinician or treating physician will know how and when to interrupt, adjust or terminate therapy in consideration of individual patient response.

Any suitable route of administration may be employed for providing the patient with an effective dosage of a R-NSAID. For example, oral, rectal, transdermal, parenteral (subcutaneous, intramuscular, intravenous), intrathecal, and like forms of administration may be employed. Dosage forms include tablets, troches, dispersions, suspensions, solutions, capsules, patches, and the like.

In order to aid in patient compliance with daily dosage requirements, the R-NSAIDs may also be administered by formulating them in a toothpaste. The drug is dissolved in an ethyl alcohol solution and added to the toothpaste so that the final concentration of R-NSAID is from about 0.01 to about 1% on a weight compositions of the present invention basis.

The present method of treatment of colorectal cancer and other neoplastic diseases will be enhanced by the use of an R-NSAID as an adjuvant to known chemotherapeutic agents such as 5-fluorouracil and the like.

The pharmaceutical compositions of the present invention comprise an R-NSAID, or a pharmaceutically acceptable salt thereof, as the active ingredient and may also contain a pharmaceutically acceptable carrier, and optionally, other therapeutic ingredients.

The terms "pharmaceutically acceptable salts" or "a pharmaceutically acceptable salt thereof" refer to salts prepared from pharmaceutically acceptable, non-toxic acids or bases. Suitable pharmaceutically acceptable salts include metallic salts, *e.g.* salts of aluminum, calcium, lithium, magnesium, potassium, sodium and zinc or organic salts, *e.g.* salts of lysine, N,N'-dibenzylethylenediamine, chloroprocaine, choline, diethanolamine,

ethylenediamine, meglumine (N-methylglucamine), procaine and tris.

The term "with reduced gastrointestinal toxicity" as used herein means that the administration of the particular R-NSAID is less ulcerogenic to the gastrointestinal tract of the human or other mammal than the corresponding racemate or S-NSAID. One measure of ulcerogenic activity is the small bowel ulcer score. A rat is treated daily through oral administration of the R-NSAID for 30 days. At the end of the 30 days, the rat is sacrificed and the intestines removed. Lesions of appreciable size in the mucosa are measured. A cumulative score equaling the sum of the diameters of the ulcers measured are reported as the ulcer score. An ulcer score essentially equal to that of a control rat, or a reduction of the ulcer score of at least 50 to 90%, preferably at least 80%, as compared to the corresponding S-NSAID or racemate, is considered a reduction in gastrointestinal toxicity.

The compositions of the present invention can be prepared in any desired form, for example, tablets, powders, capsules, suspensions, solutions, elixirs, and aerosols. Carriers such as starches, sugars, microcrystalline cellulose, diluents, granulating agents, lubricants, binders, disintegrating agents, and the like may be used in the cases of oral solid preparations. Oral solid preparations (such as powders, capsules, and tablets) are preferred over oral liquid preparations. The most preferred oral solid preparations are tablets. If desired, tablets may be coated by standard aqueous or nonaqueous techniques.

In addition to the common dosage forms set out above, the compounds of the present invention may also be administered by controlled release means and/or delivery devices such as those described in U.S. Pat. Nos.: 3,845,770; 3,916,899; 3,536,809; 3,598,123; and 4,008,719, the disclosures of which are hereby incorporated by reference in their entireties.

Pharmaceutical compositions of the present invention suitable for oral administration may be presented as discrete units such as capsules, cachets, or tablets, or aerosol sprays, each containing a predetermined amount of the active ingredient, as a powder or granules, or as a solution or a suspension in an aqueous liquid, a non-aqueous liquid, an oil-in-water emulsion, or a water-in-oil liquid emulsion. Such compositions may be prepared by any of the conventional methods of pharmacy, but all methods include the step of bringing into association the active ingredient with the carrier which constitutes one or more necessary ingredients. In general, the compositions are prepared by uniformly and

intimately admixing the active ingredient with liquid carriers or finely divided solid carriers or both, and then, if necessary, shaping the product into the desired presentation.

For example, a tablet may be prepared by compression or molding, optionally, with one or more accessory ingredients. Compressed tablets may be prepared by compressing in a suitable machine the active ingredient in a free-flowing form such as powder or granules, optionally mixed with a binder, lubricant, inert diluent, surface active or dispersing agent. Molded tablets may be made by molding, in a suitable machine, a mixture of the powdered compound moistened with an inert liquid diluent. Desirably, each tablet contains from about 0.1 mg to about 1000 mg of the active ingredient, and each cachet or capsule contains from about 0.1 mg to about 600 mg of the active ingredient. Most preferably, the tablet, cachet or capsule contains either one of four dosages, about 0.1 mg, about 50 mg, about 100 mg and about 200 mg of the active ingredient.

It has further surprisingly been discovered that the effect of R-flurbiprofen (an analog of R-ibuprofen) on neutrophil traffic is as effective as the S-enantiomer in the normal rat. Both isomers produce neutrophil adhesiveness in the post-capillary venules and extravasation of neutrophils into the surrounding tissue. Accordingly, high doses of enantiomerically stable *R*-enantiomers are believed to duplicate the salutary high dose effects of *rac*-ibuprofen in the treatment of cystic fibrosis, without the attendant COX-mediated toxicity.

Thus, in accordance with the present invention, cystic fibrosis patients are treated with R-NSAIDs at high dose, that is, at an effective cystic fibrosis therapeutic amount. As used herein, an "effective cystic fibrosis therapeutic amount" is that amount which will relieve CF symptoms which can be measured by improved pulmonary function. More specifically, a preferred effective cystic fibrosis therapeutic amount will be within the range from about 200 to 2000 mg of the selected R-NSAID per day, the amount being preferably administered in a divided dose based on the plasma half-life of the particular R-NSAID. For example, *R*-flurbiprofen is administered in 50 mg increments to achieve a target concentration of > 1 μg/mL plasma, max 500 mg (approximately 10 mg/kg body weight). The target dose is then administered bid (every 12 hours). Since the non-COX inhibiting *R*-enantiomers do not significantly effect venule caliber, none of the conventional side effects of NSAIDS occur.

Additionally, it has surprisingly been discovered that administration of an enantiomerically stable *R*-enantiomer of an NSAID appears to prevent or delay the onset of Alzheimer's Disease, again without the attendant COX-mediated toxicity.

Thus, in accordance with the present invention, patients at risk of developing Alzheimer's Disease patients are treated with R-NSAIDs at high dose, that is, at an effective Alzheimer's Disease prophylactic amount. As used herein, an "effective Alzheimer's Disease prophylactic amount" is that amount which will delay the onset of symptoms of AD by at least 6 months. More specifically, a preferred effective AD prophylactic amount will be within the range from about 50 to 2000 mg of the selected R-NSAID per day, the amount again being preferably administered in a divided dose based on the plasma half-life of the particular R-NSAID.

The invention is further illustrated by reference to the following examples describing the preparation of some of the compositions of the present invention, as well as their utility. It will be apparent to those skilled in the art that many modifications, both to materials and methods, may be practiced without departing from the purpose and interest of this invention.

EXAMPLE 1: Chemoprotective Effect and Toxicity of R-Flurbiprofen

A study was performed to compare the R- and S-isomers of flurbiprofen with regard to their effect on the Labelling Index (LI) and duodenal ulceration in contrast to the S-isomer.

Female Sprague-Dewley rats were randomized to 4 groups (N = 10) receiving 6.3 mg/Kg/day R-flurbiprofen; 6.3 mg/Kg/day S-flurbiprofen; 12.5 mg/Kg/day racemic flurbiprofen; or vehicle control. Fasted rats were sacrificed after 30 days. Small bowel ulcer score was recorded in each group.

The LI is calculated using a histologic biomarker of proliferating cells, a monoclonal antibody to Bromo-deoxyuridine (BrD-U). Intestinal crypts are examined microscopically in longitudinal sections such that proliferating cells are identified and quantified as a proportion of total crypt cells. An LI was determined for each rat using BrD-U staining to identify the proportion of mitotic cells in the crypt of Leiberkuhn. Twelve well-oriented crypts (distal colon) were examined in each rat.

The small bowel ulcer score was 0.05; 0.62; 4.54; and 3.22 in the control, R-flurbiprofen, S-flurbiprofen and racemic flurbiprofen groups, respectively. The LI was 12.62 in control animals. The LI was reduced to 8.71 and 9.09 in the R-

and S-flurbiprofen treated animals, ($P < 0.05$) and further reduced in animals receiving equal-molar doses of both enantiomers.

The results of this study indicate that R-flurbiprofen is much less ulcerogenic than its S-enantiomer, yet suppresses cell proliferation in the distal colon, a chemopreventive effect.

EXAMPLE 2: Toxicity of R-Etodolac

The effects of the isomers of etodolac in the guinea pig are determined as follows. Groups of 6-10 guinea pigs are dosed orally with either vehicle, racemic etodolac (2, 10, 5, 1 and 0.2 mg/kg), S-etodolac (20, 10, 5, 1 and 0.1 mg/kg), or R-etodolac (2, 10, 5, 1 and 0.1 mg/kg). Within 24 hours after the dose, the animals are euthanized and gross abnormalities are recorded in the GI tracts, with particular attention to the gastric mucosa of the stomach. Microerosions and redness (irritations) are noted, and the effects are compared between the treatment groups as described by Abert & Larsson (Acta Pharmacol. Toxicol. 28: 249-257, 1970). Based on such observations, the R-isomer is seen to cause virtually no gastrointestinal irritation.

EXAMPLE 3: Inhibitory Effect on the Activity of Cyclooxygenase

Cyclooxygenase inhibitors (for example aspirin and indomethacin) are known to cause damage and irritation of the gastric mucosa. Assays to determine the inhibitory effect of R-, S- and racemic ketoprofen, reference agents and vehicles on cyclooxygenase activity are conducted using RBL-1 cells (rat basophilic leukemia cell line). The effects of the test compounds, reference agents or vehicles are assessed on the cyclooxygenase-mediated production of $PGF_{2\alpha}$.

RBL-1 cells are grown in culture in Eagle's minimum essential medium supplemented with 12% fetal bovine serum and 1:100 antibiotic/antimycotic mixture at 27° C. Cells are harvested via centrifugation, washed with cold phosphate buffered saline (PBS), and suspended in PBS supplemented with 0.88 μM $CaCl_2$. Cells are incubated in the presence of a screening concentration of that compound or reference agent. Alternatively, cells are incubated in the presence of a vehicle.

Following the incubation period, cyclooxygenase activity is stimulated by the addition of 5 μM of a calcium ionophore to the incubation medium. The reaction is terminated by chilling the tubes on ice.

The cells are then separated via centrifugation, and the supernatant is removed. Aliquots of the supernatant are used to measure the calcium-ionophore-

stimulated production of $PGF_{2\alpha}$ via radioimmunoassay.

For each experiment, a vehicle-control is evaluated. A reference standard is also evaluated at a single concentration with each assay.

The results from the aforementioned studies indicate hat R-NSAIDs are safe alternatives for chemoprophylaxis in colon cancer. R-NSAIDs suppress cell proliferation in the distal colon, an anti-neoplastic effect.

What is claimed is:

1. A composition comprising an enantiomerically stable R-NSAID or a pharmaceutically acceptable salt thereof, said composition being substantially free of the S-enantiomer of said R-NSAID.

2. The composition of claim 1 further comprising a pharmaceutically acceptable carrier.

3. The componsition of claim 1 wherein said R-NSAID or salt thereof is present in an amount effective to elicit a chemoprotective effect.

4. The componsition of claim 1 wherein said R-NSAID or salt thereof is present in an amount effective against a neoplastic disease.

5. The composition of claim 1 wherein said R-NSAID or salt thereof is present in an effective cystic fibrosis therapeutic amount.

6. The composition of claim 1 wherein said R-NSAID or salt thereof is present in an effective Alzheimer's Disease prophylactic amount.

7. The composition of claims 1 wherein the ratio of said R-NSAID to said S-NSAID is at least 90:10 by weight.

8. The composition of claim 7 wherein the ratio of said R-NSAID to said S-NSAID is at least 99:1 by weight.

9. The composition of claim 1 wherein said R-NSAID is selected from the group consisting of R-flurbiprofen, R-ketoprofen, R-naproxen, R-etodolac, R-ketorolac, R-tiaprofenic acid, R-suprofen, R-carprofen, R-pirprofen, R-indoprofen, and R-benoxaprofen.

10. The composition of claim 9 wherein said R-NSAID is R-flurbiprofen.

11. The composition of claim 1 wherein said pharmaceutically acceptable salt is a metal salt or an organic salt.

12. The composition of claim 11 wherein said R-NSAID metal salt is selected from the group consisting of sodium, potassium, calcium, magnesium, lithium, aluminum and zinc salts.

13. The composition of claim 11 wherein said R-NSAID organic salt is selected from the group consisting of lysine, N,N'-dibenzylethylenediamine, chloroprocaine, choline, diethanolamine, ethylenediamine, meglumine, procaine and tris salts.

14. The composition of claim 1 comprising 0.1 mg to 2000 mg of said R-NSAID or pharmaceutically acceptable salt.

15. The composition of claim 14 comprising 0.1 mg to 600 mg of said R-NSAID or pharmaceutically acceptable salt.

16. The composition of claim 15 comprising 0.1 mg to 500 mg of said R-NSAID or pharmaceutically acceptable salt.

17. The composition of claim 1 comprising at least one further chemotherapeutic agent.

18. The composition of claim 17 wherein said chemotherapeutic agent is 5-fluorouracil.

19. A composition comprising

(a) an enantiomerically stable R-NSAID or a pharmaceutically acceptable salt thereof in an amount effective to elicit a chemoprotective effect, and

(b) a pharmaceutically acceptable carrier,

wherein the ratio of said R-NSAID to its corresponding S-enantiomer in said composition is at least 90:10 by weight.

20. A composition comprising

(a) an enantiomerically stable R-NSAID or a pharmaceutically acceptable salt thereof in an effective Alzheimer's Disease prophylactic amount, and

(b) a pharmaceutically acceptable carrier,

wherein the ratio of said R-NSAID to its corresponding S-enantiomer in said composition is at least 90:10 by weight.

21. A composition comprising

(a) 25 mg to 2000 mg of an enantiomerically stable R-NSAID selected from the group consisting of R-ketoprofen, R-flurbiprofen, R-naproxen, R-etodolac, R-ketorolac, R-tiaprofenic acid, R-suprofen, R-carprofen, R-pirprofen, R-indoprofen, and R-benoxaprofen, or a pharmaceutically acceptable salt thereof, and

(b) a pharmaceutically acceptable carrier,

wherein the ratio of said R-NSAID to its corresponding S-enantiomer in said composition is at least 90:10 by weight.

22. The composition of any of claims 19-21 wherein said ratio is at least 99:1 by weight.

23. The composition of any of claims 19-21 wherein said R-NSAID is R-flurbiprofen.

24. A method of eliciting a colorectal chemoprotective effect with reduced gastrointestinal toxicity in a mammal, comprising the step of administering to said mammal the composition of any of claims 1-3, 9-17, 21 or 23.

25 The method of claim 24 wherein said mammal is a human.

26. The method of claim 24 wherein said R-NSAID is administered orally, transdermally, intravenously or intrathecally.

27. The method of claim 24 wherein the amount of said R-NSAID or a pharmaceutically acceptable salt thereof administered is from 0.1 mg to 2000 mg per day.

28. The method of claim 27 wherein the amount administered is from 0.1 mg to 500 mg per day.

29. A method of treating a neoplastic disease with reduced gastrointestinal toxicity in a mammal, comprising the step of administering to said mammal the composition of any of claims 1, 2, 4, 9-17 or 21.

30. The method of claim 29 wherein said neoplastic disease is selected from the group consisting of breast cancer, lung cancer and prostate cancer.

31. The method of claim 29 wherein said mammal is a human.

32. The method of claim 29 wherein said R-NSAID is administered orally, transdermally, intravenously or intrathecally.

33. The method of claim 29 wherein the amount of R-NSAID administered is from 1.0 mg to 2000 mg per day.

34. A method of treating cystic fibrosis comprising the step of administering to a patient in need of said treatment the composition of any of claims 1, 2, 5, 9-17 or 21.

35. The method of claim 34 wherein said composition comprises about 1 mg to 600 mg of said R-NSAID or salt.

36. The method of claim 34 wherein said R-NSAID is administered orally, transdermally, intravenously or intrathecally.

37. A method of preventing or delaying the onset of symptoms of Alzheimer's Disease comprising the step of administering to a patient in need of said treatment the composition of any of claims 1, 2, 6, 9-17, 20 or 21.

38. The method of claim 37 wherein said composition comprises about 1 mg to 600 mg of said R-NSAID or salt.

39. The method of claim 37 wherein said R-NSAID is administered orally, transdermally, intravenously or intrathecally.

40. Use of an enantiomerically stable R-NSAID or a pharmaceutically acceptable salt thereof in preparing a medicament for use in eliciting a colorectal chemoprotective effect with reduced gastrointestinal toxicity in a mammal, said medicament being substantially free of the S-enantiomer of said R-NSAID.

41. Use of an enantiomerically stable R-NSAID or a pharmaceutically acceptable salt thereof in preparing a medicament for use in treating a neoplastic disease with reduced gastrointestinal toxicity in a mammal, said medicament being substantially free of the S-enantiomer of said R-NSAID.

42. Use of an enantiomerically stable R-NSAID or a pharmaceutically acceptable salt thereof in preparing a medicament for use in treating cystic fibrosis, said medicament being substantially free of the S-enantiomer of said R-NSAID.

43. Use of an enantiomerically stable R-NSAID or a pharmaceutically acceptable salt thereof in preparing a medicament for use in preventing or delaying the onset of symptoms of Alzheimer's Disease, said medicament being substantially free of the S-enantiomer of said R-NSAID.